[0234] A 500 mg portion of compound (**DJ**) (produced in accordance with Example 67 and illustrated in Figure 33) was dissolved in 5 mL of dry THF. The mixture was treated with 1.002 mL (10 equivalents) of ethylenediamine (EDA), and allowed to stir for 2 hours. The solution was decanted from the formed solid. The solvent and excess EDA were removed from the decanted solution by rotoevaporation under vacuum to yield 0.8728 g of white solid. The product (**DN**) was characterized by LCMS: retention time 1.608 minutes and desired M+H observed at 294 *m/z*.

EXAMPLE 46

[0235] This example provides a method of generating compound (**CS**) in Figure 33.

[0236] A solution of 872 mg of the amine prepared in Example 45 in 10 mL dioxane was treated with paraformaldehyde (535 mg) and trimethylphosphite (2.21 g). The mixture was heated at 100° C overnight. The solvent was removed by rotary evaporation at 80° C to give a brown solid. Chloroform (25 mL) was added, and the solution was washed with water (15 mL). The organics were dried over sodium sulfate, and the solvent was removed to provide 241 mg of a yellow semi-solid. The resulting solid was purified via LC to provide 58.8 mg of compound (**CS**). The presence of compound (**CS**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 2.6 minutes, MS [M=$C_{21}H_{37}N_3O_9P_2$] *m/z* 538 (MH^+), 560 (MNa^+).

EXAMPLE 47

[0237] This example provides a method of generating compound (**CT**) in Figure 33.

[0238] A solution of 54.6 mg of the phosphonate prepared in Example 46 in 1 mL dichloromethane was treated with bromotrimethylsilane (156 mg). The mixture was stirred overnight. Ethanol (0.5 mL) and water (3 drops) were added, and the mixture was stirred for 1 hour. The volatiles were removed, and the material was dried under vacuum. The resulting material was taken up in water (1 mL) and lyophilized to provide 59 mg of a tan solid. The presence of compound (**CT**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 0.4 minutes, MS [M=$C_{12}H_{21}N_3O_7P_2$] *m/z* 380 ($M-H^-$), 382 (MH^+), 404 (MNa^+).

EXAMPLE 48

[0239] This example provides a method of generating compound (**CU**) in Figure 33.

[0240] A mixture of 50 mg pifithrin-α hydrobromide, mono-methyl terephthalate (1.0 equivalents), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (1.5 equivalents), 1-hydroxybenzotriazole (1.5 equivalents), and diisopropylethylamine (8.0 equivalents) in 2 mL dimethylformamide was stirred overnight. Water (5 mL) was added,

producing a precipitate, which was filtered and washed with water (3 x 2 mL). The material was dried under vacuum to provide 47.7 mg of a yellow solid. The presence of compound (**CU**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 4.6 minutes, MS [M=$C_{25}H_{24}N_2O_4S$] *m/z* 447 ($M\text{-}H^-$), 449 (MH^+).

EXAMPLE 49

[0241] This example provides a method of generating compound (**CV**) in Figure 33.

[0242] A solution of 40 mg of pifithrin-β-CH_2Cl (prepared according to the procedure of Example 23) in 2 mL acetonitrile was treated with mono-*tert*-butyl succinate (3.0 equivalents) and diisopropylethylamine (3 equivalents). The mixture was stirred overnight. The solvent was removed, and the material was purified via LC to provide 22 mg of desired product. The presence of compound (**CV**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 4.3 minutes, MS [M=$C_{25}H_{31}N_2O_4S$] *m/z* 430(M^+).

EXAMPLE 50

[0243] This example provides a method of generating compound (**CW**) in Figure 33.

[0244] A solution of 40 mg pifithrin-β in 800 μL acetonitrile was treated with oxalyl chloride (2.0 equivalents) and was stirred overnight. Three drops of this mixture was added to a vial containing benzylamine (1.5 μL) in acetonitrile (500 μL). The presence of compound (**CW**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 2.8 minutes, MS [M=$C_{25}H_{24}N_3O_2S$] *m/z* 455(M^+).

EXAMPLE 51

[0245] This example provides a method of generating compound (**CX**) in Figure 33.

[0246] A solution of 40 mg pifithrin-β in 800 μL acetonitrile was treated with oxalyl chloride (2.0 equivalents) and was stirred overnight. Three drops of this mixture was added to a vial containing methanol (500 μL). The presence of compound (**CX**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 4.4 minutes, MS [M=$C_{19}H_{19}N_2O_3S$] *m/z* 355(M^+), 396 ($M\text{-}CH_3CN^+$).

EXAMPLE 52

[0247] This example provides a method of generating compound (**CY**) in Figure 33.

[0248] Compound (**CY**) was prepared via the procedure of Kantoci et al., *Syn. Commun.*, *26*(10), 2037 (1996). A mixture of N-benzyl-N-methylamine (20.0 g), diethylphosphite (70.7 g) and triethylorthoformate (29.3 g) was stirred under argon at reflux (150° C) for 5 hours. Ethanol was removed via rotary evaporation at 70° C, and the mixture

was again heated at reflux overnight. The solution was diluted with 600 mL chloroform, and was washed with 1 M sodium hydroxide (3 x 100 mL) and saturated sodium chloride (3 x 150 mL). The organics were dried over sodium sulfate, and the solvent was removed to provide 74.0 g of a light yellow oil. Ten grams of this material was subjected to silica gel column chromatography using 14:4:1 ethyl acetate: hexane: methanol as eluent. This provided 6.08 g of a clear oil. The presence of compound (**CY**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 3.4 minutes, MS [M=$C_{17}H_{31}NO_6P_2$] *m/z* 408(MH^+), 430 (MNa^+), 471 ($MNa\text{-}CH_3CN^+$).

EXAMPLE 53

[0249] This example provides a method of generating compound (**CZ**) in Figure 34.

[0250] Compound (**CZ**) (CAS # 80475-00-9) was prepared via the procedure of Kantoci et al., *supra*. A solution of 4.52 g of the phosphonated benzylamine prepared in Example 52 in methanol (45 mL) was treated with 10% palladium on carbon (200 mg) and subjected to an atmosphere of hydrogen overnight. The palladium/carbon was filtered to provide 2.98 g of a light yellow oil. The presence of compound (**CZ**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 1.9 minutes, MS [M=$C_{10}H_{25}NO_6P_2$] *m/z* 318(MH^+).

EXAMPLE 54

[0251] This example provides a method of generating compound (**DA**) in Figure 34.

[0252] A solution of 40 mg pifithrin-β in 800 μL acetonitrile was treated with oxalyl chloride (2.0 equivalents) and was stirred overnight. To the solution was added the aminophosphonate prepared in Example 53 (3 equivalents). The presence of compound (**DA**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 4.0 minutes, MS [M=$C_{28}H_{40}N_3O_8P_2S$] *m/z* 640(M^+).

EXAMPLE 55

[0253] This example provides a method of generating compound (**DB**) in Figure 34.

[0254] A mixture of 100 mg pifithrin-α hydrobromide, terephthalic acid (1.0 equivalents), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (2.0 equivalents), 1-hydroxybenzotriazole (2.0 equivalents), and diisopropylethylamine (10.0 equivalents) in 4 mL dimethylformamide was stirred overnight. Water (11 mL) was added and the mixture was centrifuged, removing the solid. The resulting liquid was diluted in dichloromethane (30 mL) and 1 M hydrochloric acid (30 mL). The layers were separated, and the aqueous layer was extracted with fresh dichloromethane (30 mL). The organics were dried over sodium sulfate, and the residue was purified via LC to give 11.3 mg of a

yellow solid. The presence of compound (**DB**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 4.0 minutes, MS [M=$C_{24}H_{22}N_2O_4S$] *m/z* 433 ($M\text{-}H^-$), 435 (MH^+).

EXAMPLE 56

[0255] This example provides a method of generating compound (**DC**) in Figure 34.

[0256] A solution of 50 mg pifithrin-α free base prepared in Example 44 in dichloromethane (2 mL) was treated with oxalyl chloride (1.5 equivalents). After 1 hour, a solution of benzylamine (4.0 equivalents) in dichloromethane (1 mL) was added. The mixture was stirred overnight. The presence of compound (**DC**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 4.1 minutes, MS [M=$C_{25}H_{25}N_3O_3S$] *m/z* 448 (MH^+).

EXAMPLE 57

[0257] This example describes a method of generating compound (**DO**) of Figure 36.

[0258] A 500 mg sample of 4-chloromethylbenzoic acid (Aldrich) was dissolved in 8 mL of THF and treated all at once with 5 equivalents of ethylenediamine (Aldrich) (983 uL). After 24 hours, the solvent was removed under high vacuum. The resulting white solid (93%), compound (**DO**), was characterized by LCMS: retention time 0.4 minutes, desired M+H observed at 195 *m/z*.

EXAMPLE 58

[0259] This example provides a method of generating compound (**DD**) in Figure 34.

[0260] A mixture of 679 mg of the amino acid prepared in Example 57, 37% (wt/wt) aqueous formaldehyde (1.04 mL), phosphorus acid (1.15 g), and concentrated (12.1 M) hydrochloric acid (2.3 mL) in dioxane (10 mL) was stirred at 100° C overnight. The solvent was removed via rotary evaporation at 75° C. The mixture was centrifuged and the solid discarded. To the liquid was added more formaldehyde solution (1.04 mL), phosphorus acid (1.15 g), concentrated hydrochloric acid (2 mL), and dioxane (10 mL). The mixture was stirred overnight at 100° C. The solvent was removed via rotary evaporation at 75° C to provide a thick oil. This was purified via LC to provide 276 mg of a brown solid. The presence of compound (**DD**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 0.6 minutes, MS [M=$C_{13}H_{23}N_2O_{11}P_3$] *m/z* 475 ($M\text{-}H^-$), 477 (MH^+).

EXAMPLE 59

[0261] This example provides a method of generating compound (**DE**) in Figure 34.

[0262] A solution of 500 mg pifithrin-α hydrobromide in acetonitrile (10 mL) was treated with di-*tert*-butyldicarbonate (1.1 equivalents) and triethylamine (2.1 equivalents). The mixture was stirred overnight, and the solvent was removed. Dichloromethane (20 mL) was added, and the solution was washed with water (10 mL). The organics were dried over sodium sulfate, and the solvent was removed to provide 488 mg of a tan solid. The presence of compound (**DE**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 4.5 minutes, MS [M=$C_{21}H_{26}N_2O_3S$] *m/z* 387 (MH^+), 409 (MNa^+), 450($MNa\text{-}CH_3CN^+$).

[0263] The BOC protecting group in compound DE allows for modification of the ketone functionality as illustrated by the preparation of compound DF in Example 62. Other conversions of the ketone functionality of compound DE can be accomplished using any conditions that do not remove the BOC protecting group (i.e. strong acids will remove the protecting group). After the conversion of the ketone into a reversible linker (as exemplified by DF in Example 62) the BOC protecting group can then be removed under acidic conditions (i.e. trifluoroacetic acid) or trimethylsilyl bromide to give pifithrin-alpha derivatives reversibly substituted only on the ketone group that are not readily obtainable by other methods.

EXAMPLE 60

[0264] This example provides a method of generating compound (**DF**) in Figure 34.

[0265] A solution of 100 mg of BOC-protected pifithrin-α prepared in Example 59 in acetonitrile (5 mL) was treated with phenylhydrazine (3 equivalents) and a catalytic amount of *p*-toluenesulfonic acid. The mixture was stirred overnight. Dichloromethane (20 mL) was added, and the solution was washed with water (20 mL). The organics were dried over sodium sulfate, and the solvent was removed to provide 131 mg of a brown oil. The presence of compound (**DF**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 5.8 minutes, MS [M=$C_{27}H_{32}N_4O_2S$] *m/z* 499 (MNa^+), 540($MNa\text{-}CH_3CN^+$).

EXAMPLE 61

[0266] This example provides a method of generating compound (**DG**) in Figure 34.

[0267] A mixture of 6.0 g Fmoc-Lys-OH (Advanced ChemTech) in methanol (25 mL) and water (25 mL) was treated with 37% (wt/wt) aqueous formaldehyde (6.06 mL) and dimethylphosphite (8.96 g). The mixture was stirred at 80° C for 2 hours, cooled, and extracted with dichloromethane (1 x 100 mL, 2 x 50 mL). The organics were washed with saturated sodium chloride (50 mL), dried over magnesium sulfate for 30 minutes, and the solvent was removed to provide 10.17 g of a light green oil. The presence of compound

(**DG**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 3.3 minutes, MS [M=$C_{27}H_{38}N_2O_{10}P_2$] *m/z* 613 (MH^+), 635 (MNa^+).

EXAMPLE 62

[0268] This example provides a method of generating compound (**DH**) in Figure 35.

[0269] A mixture of 83 mg pifithrin-α free base prepared in Example 44, phosphonated Fmoc-Lys-OH prepared in Example 61 (1.1 equivalents), 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (1.5 equivalents), 1-hydroxybenzotriazole (1.5 equivalents), and diisopropylethylamine (6.0 equivalents) in 2 mL dimethylformamide was stirred overnight. The presence of compound (**DH**) was confirmed by electrospray LC-MS; t_R = 4.4 minutes, MS [M=$C_{43}H_{54}N_4O_{10}P_2S$] *m/z* 881 ($MH^+$), 903 ($MNa^+$).

EXAMPLE 63

[0270] This example provides a method of generating compound (**DI**) in Figure 35.

[0271] A solution of 23.9 mg phosphonated Fmoc-Lys-OH prepared in Example 61 in dichloromethane (1 mL) was treated with bromotrimethylsilane (60 mg). The mixture was stirred overnight. The presence of compound (**DI**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 3.4 minutes, MS [M=$C_{23}H_{30}N_2O_{10}P_2$] *m/z* 555 ($M-H^-$).

EXAMPLE 64

[0272] This example provides an alternate method of generating compound (**DI**) in Figure 35.

[0273] A solution of 112.9 mg phosphonated Fmoc-Lys-OH prepared in Example 61 in 6 M hydrochloric acid (3 mL) was stirred at 80° C for 2 days. Water (9 mL) was added. After 2 more days, the mixture was centrifuged and the liquid decanted. The solid was dried under vacuum to provide 86.8 mg of an off-white solid. The presence of compound (**DI**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 3.1 minutes, MS [M=$C_{23}H_{30}N_2O_{10}P_2$] *m/z* 555 ($M-H^-$).

EXAMPLE 65

[0274] This example provides an alternate method of generating compound (**DI**) in Figure 35.

[0275] A solution of 500 mg Fmoc-Lys-OH (Advanced ChemTech) in dioxane (5 mL) was treated with 37% (wt/wt) aqueous formaldehyde (303 μL), phosphorus acid (333 mg), and concentrated (12.1 M) hydrochloric acid (674 μL). The mixture was stirred at 90° C overnight, and the solvent was removed via rotary evaporation at 75° C. The presence of

compound (**DI**) was confirmed by electrospray LC-MS; t_R = 5.4 minutes, MS [M=$C_{23}H_{30}N_2O_{10}P_2$] *m/z* 555 ($M-H^-$).

EXAMPLE 66

[0276] The example provides a method of producing compound (**DJ**) of Figure 36.

[0277] A 1.5 g portion of compound (**DP**) of Example 71 was dissolved in 17 mL of dry THF along with 0.69 g of N-hydroxysuccinimide (Aldrich). The mixture was treated all at once with 6 mL of 1M dicyclohexylcarbodiimide in dichloromethane with stirring. After 2 days, a white precipitate (dicyclohexylurea) was filtered. The filtrate was rotoevaporated under vacuum to yield 2.8146 g of white solid, which was characterized by LCMS: retention time 3.299 minutes and desired M+H observed at 349 *m/z*.

EXAMPLE 67

[0278] This example provides a method of generating compound (**DJ**) in Figure 35.

[0279] A solution of 1.29 g of the Boc-protected amino acid prepared in Example 71 in 15 mL tetrahydrofuran was treated with N-hydroxysuccinimde (623 mg) and 1.0 M 1,3-dicyclohexylcarbodiimide in dichloromethane (5.4 mL). The mixture was stirred overnight. A white precipitate was filtered, and the supernatant was concentrated to provide 1.89 g of a white solid. . The presence of compound (**DJ**) was indicated by the presence of a UV signal associated with the peak at 3.3 minutes.

EXAMPLE 68

[0280] This example provides a method of generating compound (**DK**) in Figure 35.

[0281] To a solution of 2.1 g tris-(2-aminoethyl)amine in 20 mL tetrahydrofuran, a solution of 1.0 g of the activated ester prepared as in Example 66 in 20 mL tetrahydrofuran was added drop-wise over a period of 40 minutes. The mixture was stirred overnight resulting in a precipitate that was filtered and concentrated via rotary evaporation to provide 2.10 g of a yellow oil. The presence of compound (**DK**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 1.4 minutes, MS [M=$C_{19}H_{33}N_5O_3$] *m/z* 380 (MH^+), 402 (MNa^+).

EXAMPLE 69

[0282] This example provides a method of generating compound (**DL**) in Figure 35.

[0283] A solution of 2.08 g of the amine prepared in Example 68 in dioxane (20 mL) was treated with paraformaldehyde (1.50 g) and dimethylphosphite (6.85 g). The mixture was stirred at 90° C overnight, and the solvent was removed via rotary evaporation at 70° C. Dichloromethane (50 mL) was added, and the mixture was washed with saturated sodium

chloride (25 mL) and water (25 mL). The organics were dried over sodium sulfate, and the solvent was removed. The residue was purified via LC to provide 123.8 mg of a yellow oil. The presence of compound (**DL**) was confirmed by electrospray LC-MS and exhibited the following characteristics: t_R = 2.2 minutes, MS [M=$C_{31}H_{61}N_5O_{15}P_4$] *m/z* 868 (MH^+).

EXAMPLE 70

[0284] This example provides a method of generating compound (**DM**) in Figure 35.

[0285] A solution of 111.1 mg of the phosphonated diamine prepared in Example 69 in 1 mL dichloromethane was treated with 194 mg of bromotrimethylsilane. After 5 hours, methanol (1 mL) was added. The mixture was stirred for 1 hour, and the solvent was removed to provide 113.9 mg of a tan solid. The presence of compound (**DM**) was confirmed by electrospray LC-MS and exhibited the following characteristics; t_R = 1.0 minutes, MS [M=$C_{18}H_{37}N_5O_{13}P_4$] *m/z* 328 [$(M+2H/2)^{2+}$)], 656 (MH^+). The compound also was analyzed by proton NMR spectroscopy: ^1H ($CDCl_3$) δ: 7.77 (d, 2H, *J* 8.1 Hz), 7.43 (d, 2H, *J* 8.2 Hz), 4.1-3.3 (m, 33H).

EXAMPLE 71

[0286] This example provides a method of generating compound (**DP**) in Figure 36.

[0287] A 2.0 g portion of 4-aminomethyl benzoic acid (Aldrich) was dissolved in 20 mL of water containing 0.64 g of solid NaOH. A 3.18 g portion of Boc anhydride (Aldrich) was added, and the resulting mixture was stirred overnight. The mixture was adjusted to pH=2 by the careful addition of 15 mL of 2N HCl. The resulting white solid was filtered and dried to yield 2.9997 g of product (**DP**). The compound (**DP**) was characterized by LCMS: retention time 2.901 minutes, desired M-H mass ion observed at 250 *m/z*.

[0288] All references, including publications, patent applications, and patents, cited herein are hereby incorporated by reference to the same extent as if each reference were individually and specifically indicated to be incorporated by reference and were set forth in its entirety herein.

[0289] The use of the terms "a" and "an" and "the" and similar referents in the context of describing the invention (especially in the context of the following claims) are to be construed to cover both the singular and the plural, unless otherwise indicated herein or clearly contradicted by context. The terms "comprising," "having," "including," and "containing" are to be construed as open-ended terms (i.e., meaning "including, but not limited to,") unless otherwise noted. Recitation of ranges of values herein are merely intended to serve as a shorthand method of referring individually to each separate value

falling within the range, unless otherwise indicated herein, and each separate value is incorporated into the specification as if it were individually recited herein. All methods described herein can be performed in any suitable order unless otherwise indicated herein or otherwise clearly contradicted by context. The use of any and all examples, or exemplary language (e.g., "such as") provided herein, is intended merely to better illuminate the invention and does not pose a limitation on the scope of the invention unless otherwise claimed. No language in the specification should be construed as indicating any non-claimed element as essential to the practice of the invention.

[0290] Preferred embodiments of this invention are described herein, including the best mode known to the inventors for carrying out the invention. Variations of those preferred embodiments may become apparent to those of ordinary skill in the art upon reading the foregoing description. The inventors expect skilled artisans to employ such variations as appropriate, and the inventors intend for the invention to be practiced otherwise than as specifically described herein. Accordingly, this invention includes all modifications and equivalents of the subject matter recited in the claims appended hereto as permitted by applicable law. Moreover, any combination of the above-described elements in all possible variations thereof is encompassed by the invention unless otherwise indicated herein or otherwise clearly contradicted by context.